Numb, and a similar gene Numblike, are expressed in activated satellite cells. This study is focused on determining the role of Numb in muscle repair. Our data shows that Numb protein isoforms demonstrate a dynamic pattern of expression during myogenesis in C2C12, and murine satellite cells. MicroRNA knockdown of Numb in C2C12 cells demonstrated up-regulation of myogenesis genes. Further, mice with conditional Numb/Numblike alleles demonstrate impaired muscle regeneration in response to induced damage.

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Program/Abstract # 381
Homology of Tβ-4 and VEGF in *Xenopus*, axolotl, and short-toes
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Thymosin beta-4 (Tβ-4) and vascular endothelial growth factor (VEGF) are important growth factors in angiogenesis, a process critical to regeneration. Axolotl (*Ambystoma mexicanum*) is a unique animal model capable of regenerating their tissue parts after amputation, while short-toes (a mutant axolotl) and African clawed frog (*Xenopus laevis*) are considered as regeneration-deficient since they cannot replace their lost parts. However, we lack sequence information for VEGF from axolotl and short-toes, and that of Tβ-4 from all three animals, which has impaired our studies of their roles in regeneration. In this study, we partially sequenced and compared the mRNA of VEGF and Tβ-4 in Axolotl, short-toes, and African clawed frog. VEGF primers were designed based on *Xenopus* cDNA sequence, while the Tβ-4 primers were designed based on human cDNA sequences. The total RNA was extracted utilizing RNeasy kit (Qiagen Sciences Inc., Germantown, MD) following the manufacturer's instruction. One-step reverse transcriptase polymerase chain reaction (RT-PCR, Qiagen) was performed following the manufacturer's instructions. The RT-PCR products were run on 0.9% agarose gel and then sent for two direction sequencing analyses (ACGT, Inc., Wheeling, IL). After comparing the RT-PCR products sequences, the result suggested that VEGF and Tβ-4 are highly conserved (>90%) among *Xenopus laevis*, axolotl and short-toes. This project is supported by the Indiana University School of Dentistry start-up funds to F. Song and by the W.M. Keck foundation to D. Stocum.

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Program/Abstract # 383
H,K-ATPase-mediated ion transport regulates anterior patterning in regenerating planaria
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To translate regeneration research into therapies will require knowledge of not just stem cell-derived proliferation but also of the subsequent patterning mechanisms. In particular, it is critical to elucidate how the newly formed tissue (blastema) communicates and integrates with existing tissues to maintain proper morphogenesis and polarity. Using the powerful planarian regeneration model, we have uncovered a role for ion transport (a known regulator of cell behavior and morphogenesis) in anterior–posterior (A/P) patterning during regeneration. Our data reveal that one key bioelectric signal controlling regeneration is the H,K-ATPase—an ion pump with roles in embryonic left–right patterning and stomach pH in vertebrates. Inactivating H,K-ATPase activity genetically (via RNAi) or chemically results in normal regeneration of the tail but a morphologically deficient anterior regenerate. Marker analyses reveal that H,K-ATPase inhibition does not duplicate posterior structures, suggesting that H,K-ATPase activity plays an important role in determining anterior axial identity in the blastema. Wounding of a pharmacologically-induced no-headed regenerate surprisingly restarts head formation if a new blastema is formed, suggesting that H,K-ATPase deficient worms retain anterior competency. Most importantly, pharmacological studies reveal that H,K-ATPase activity is essential for the mechanism by which the blastema senses distant polarity cues along the A/P axis (and across existing tissues). Together, our data uncover a novel patterning role for the hydrogen–potassium exchanger and biophysical regulation in regeneration.

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Program/Abstract # 384
Long-range neural and gap junction protein-mediated cues control polarity during planarian regeneration
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Having the ability to coordinate the behavior of stem cells to induce regeneration of specific large-scale structures would have far reaching consequences in the treatment of degenerative diseases, acute injury, and aging. Thus, identifying and learning to manipulate the sequential steps that determine the fate of new tissue within the overall morphogenetic program of the organism is fundamental. We identified novel early signals, mediated by the central nervous system and 3 innexin proteins, which determine the fate and axial polarity of regenerated tissue in planarians. Modulation of gap junction-dependent and neural signals specifically induces ectopic anterior regeneration blastemas in posterior and lateral wounds. These ectopic anterior blastemas differentiate new brains that establish permanent primary axes re-established during subsequent rounds of unperturbed regeneration. These data reveal powerful novel controls of pattern formation and suggest a constructive model linking nervous inputs and polarity determination in early stages of regeneration.

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Program/Abstract # 385
Combinational differentiation by environmental manipulation and transgene with NIA ES cell bank
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One of the critical issues in regenerative medicine is how to differentiate embryonic stem (ES) cells or induced pluripotent stem cells into the desirable cell types. To induce a specific cell lineage, two major approaches have been developed that are changing culture